

Genotoxicity and other relevant data

IARC reported there is strong evidence that glyphosate and commercial formulations can be genotoxic and produce oxidative damage. This is in stark contrast to what regulatory authorities (e.g., US EPA, Canada PMRA, EU, Japan), scientific bodies (JMPR/WHO, WHO IPCS, WHO Water) and third party experts (Williams et al., 2000; Mink et al., 2012; Kier and Kirkland, 2013; Kier 2015; and Greim et al., 2015) around the world for decades have concluded - that glyphosate is not genotoxic or carcinogenic.

The only way IARC could have come to an opinion so completely opposite regarding glyphosate and glyphosate-based formulations was to disregard a plethora of more relevant data and the opinions of numerous other scientists who have carefully considered all the available data. Further, IARC interpreted findings differently, and their review suffered from relying on non-standard studies with adverse effects which used methods that have not been validated and/or not conducted according to international guidelines. IARC also put totally inappropriate emphasis on studies that are not relevant for humans based on exposure conditions. It appears they did not conduct a well-reasoned weight-of-evidence evaluation or follow standard practice and frameworks that are the foundation of hazard and risk assessment (Adami et al., 2011 and Lewis et al., 2002)

There is an expansive data-base with studies conducted by several glyphosate registrants for regulatory purposes as well as those reported by other scientists in the open literature that assess the genotoxicity potential of glyphosate and glyphosate-based formulations. As noted above, the clear conclusion by all regulatory agencies globally (most recently by the German BfR 2015) and third party scientists (most recently Kier and Kirkland 2013 and Kier 2015) over decades looking at all the scientific data is that glyphosate and glyphosate-based products are not genotoxic.

IARC has clearly focused on the studies reporting adverse findings. These studies are compromised by various deficiencies such as: not conducted according to internationally recognized guidelines, conducted at abnormally high-dose levels, *in vitro* studies using inappropriately high concentrations of test materials, or use of irrelevant routes of exposure for humans.

Genotoxicity

While IARC has not provided specific references for all the studies they used to support their faulty conclusions, many can be deduced based on our knowledge of the database and publications in the open literature.

Two studies that have been used to make claims of genotoxicity/oxidative damage in the past are those of Bolognesi et al, 1997 and Peluso et al. 1998.

Peluso et al., directly injected test material into the abdomens (intraperitoneal or ip administration) of mice at near-lethal doses. When mice were injected with a glyphosate-based formulation which contained a surfactant, they reported what they described as evidence for DNA adducts in the kidneys and livers of these animals.

Williams et al., 2000 reviewed this study and identified a number of problems with the

procedure that led to erroneous conclusions. First, there is no evidence for a dose response over the narrow range of doses examined. Second, the level of adducts reported is so low that it is well within the range reported for normal endogenous adducts. In addition, Peluso et al., were unable to provide any chemical characterization of the product(s) that they identified as adducts, and it should be concluded that the observations of Peluso et al., are not supportive of a biologically relevant response. The route of administration is unusual, since injections of an herbicide into the abdomen is not a relevant route of exposure for humans. *[NOTE – for reporters, we can just mention this last one and identify it as the most important problem]*

Bolognesi et al, 1997 reported that ip injection of mice with glyphosate and a glyphosate-based formulation could result in DNA damage in kidney and liver. Williams et al., 2000 also reviewed this study and concluded there are several reasons to question the results from this assay. Most notably, as was the case in the Pelusa et al. study, the effects reported were only observed at doses close to or in excess of the ip LD50 for mice. Again, effects observed only at a near-lethal dose level using an irrelevant route of exposure do not demonstrate any significant risk to humans.

Heydens et al, 2008 conducted a series of mode-of-action investigations to understand the results of the Peluso et al., and Bolognesi et al., studies. The authors demonstrated that exposure by ip injection produced marked liver and kidney toxicity, but oral administration did not. The results suggest that high-dose ip injections of a formulated product may induce secondary effects mediated by local toxicity rather than genotoxicity. The large increases in 8-OHdG (a marker of oxidative stress) reported by Bolognesi et al., were not reproduced by Heydens et al. Because of the more robust nature of the Heyden et al. investigation, results of the Bolognesi et al., study do not appear to provide sufficient evidence to conclude that high-dose ip administration of glyphosate causes oxidative damage to DNA. Heydens et al. concluded that their results continue to support the conclusion that glyphosate and glyphosate-based products are not genotoxic under exposure conditions that are relevant to animals and humans.

Recently Henderson et al., 2015 concluded that in contrast to the current models their data suggests that oxidative stress is not a key determinant in the mechanism of non-genotoxic carcinogenesis.

Furthermore, Levine et al., 2007 demonstrated that surfactants can elicit cytotoxic effects such as perturbation of the mitochondrial membrane and disruption of mitochondrial membrane potential in cultured mammalian cells. The German BfR concluded surfactant effects provide a plausible mechanism for observations of glyphosate based formulations inducing DNA damage responses. Such responses would be expected to be associated with cytotoxicity-inducing exposures and exhibit a threshold.

The following statement is in reference to the Bolognesi et al., 2009 study – “One study reported increases in blood markers of chromosomal damage (micronuclei) in residents of several communities after spraying glyphosate formulations”

This is a study that attempted to evaluate possible DNA damage in people living near areas where glyphosate was used aerially to eradicate illicit crops. It is extremely difficult to draw meaningful conclusions from this study because there were so many uncontrolled variables and problems that come

with such a study, most notably, self-reporting (inaccuracies and/or information can't be verified).

There are various inconsistencies in this study that raise significant questions; for example:

- The degree of DNA damage observed immediately after the glyphosate spraying was not consistent with the application rates used,
- There was no association between self-reported direct contact with eradication sprays and DNA damage in two sprayed regions,
- The largest increase in DNA damage was reported in the region where only 1 of 25 people from this population self-reported contact with spray exposure.

The clear lack of correlations led the study authors themselves to be cautious with drawing conclusions; they made somewhat different conclusions in different places in the publication:

- In the Abstract, they say the data suggest that damage is small and appears to be transient; the evidence indicates that the genotoxic risk is low.
- In the Discussion, they also conclude that genotoxic damage is small and transient, and that genotoxic risk is of low biological relevance.

A more defensible conclusion that appears to be supported by the self-reported exposure information is that this study does not clearly demonstrate an association between glyphosate exposure and the endpoint.

A key feature overlooked by IARC is glyphosate has no structural alerts as to indicate that it would be carcinogenic or mutagenic. The Structural Alerts are molecular substructures or reactive groups that are related to the carcinogenic and mutagenic properties of the chemicals, and represent a sort of “codification” of a long series of studies aimed at highlighting the mechanisms of action of the mutagenic and carcinogenic chemicals. The identification of the Structural Alerts has had a great value both in terms of understanding mechanisms, and of assessing the risk posed by chemicals (Benigni and Cecilia Bossa, 2006).

References

Henderson CJ, Cameron AR, Chatham L, Stanley LA and Wolf CR. (2015). Evidence that the capacity of non-genotoxic carcinogens to induce oxidative stress is subject to marked variability. ToxSci Advance Access <http://toxsci.oxfordjournals.org/> at Society of Toxicology on March 18, 2015

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